Effect of trace elements on growth of *Pinus tabulaeformis* seedling

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Abstract: The stimulative effect of trace elements on seed germination and seedling growth of *Pinus tabulaeformis* was tested. The experiments were carried out on seed soak and topdressing with different trace elements and varied concentrations at the nursery of Gardens Research Institute, Harbin, in 2000-2001. The experimental results showed that soaking seed with 1% and 0.2% concentrations of Mn element produced best result for seed germination, and the germination rate was increased by 9%~19% for the seeds treated with 1% concentration and 12%~14% for the seeds treated with 0.2% concentration compared with the control group. The seeds treated with boron element had lowest germination rate. For trace element topdressing, Mn and Mo elements presented good result for seedling growth and the treatment with low concentration was even better. The height or chlorophyll content of the seedlings with spray of low-concentration Mn and Mo element was much higher than that of untreated ones. In the contrast to the treating method of seed soak, topdressing (application of spraying on foliage) had evident effect on seedling growth.

Keywords: *Pinus tabulaeformis*; Trace element; Seed soaking; Topdressing; Seed germination; Seedling growth
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Introduction

Pinus tabulaeformis is a fast-growing tree species, originally growing in north China (Zhou 1986). Some trees can survive for thousand years. It has high ornamental value and takes an important position in afforestation in northern region of China. Trace element is important component of nutrient substance of plant. Although the content of trace elements in plant is very low, it plays important role in adjustment of physiology/biochemistry as well as enzyme activities of plant (Chen 2002). Many studies have been carried out on the effect of trace element on growth and development of plants (Zhong *et al.* 1994; Fu *et al.* 1995; Hai *et al.* Tang 1998; 1999; Li *et al.* 999), but no report was found on the effect of trace elements on growth dynamic of *Pinus tabulaeformis*. This paper studied the stimulative effect of trace elements on seed germination and seedling growth of *Pinus tabulaeformis*, by soaking seed in different trace elements with varied concentrations and spraying trace element fertilizer of different concentrations on seedling foliage, with a purpose of providing an important support for improving breeding technique of the species.

Experimental material and methods

Experimental materials

The experiment was carried out at the nursery of Gardens Research Institute, Harbin. The producing area of seeds of *Pinus tabulaeformis* was southern Liaoning Province. The weight per 1000 seeds was 42.75 g, seed purity 97%, and germination rate 87%. The content of each kind of elements in soil of experimental site was shown in Table 1 and medicine used was shown in Table 2.

Table 1. Contents of every kind of element in soil of experimental site (%)

Elements	Percentage /%	Elements	Percentage/ %
N	0.27	Zn	0.0037
P	0.14	Mg	0.0037
K	0.107	Pb	0.0098
Cu	0.0024	Co	0.0002
Mn	0.009	Ni	0.0023

Table 2. Medicine used in experiment

Medicine name	Molecular formula	Symbol
Bluestone	$CuSO_4 \cdot 5H_2O$	Cu
Boracic acid	H_3BO_3	B
Manganese chloride	$MnCl_4 \cdot 4H_2O$	Mn
Sodium Molybdenum acid	$Na_2MoO_4 \cdot 7H_2O$	Mo
zinc sulfate	$ZnSO_4 \cdot 7H_2O$	Zn

Note: purity of each medicine is 97%

Experimental methods

Seed germination: Seed germination of Pinus tabulaeformis, was test on April 28, 2000 and January 3, 2001 by one-hundred-seed germination method. Three concentrations (0.02%, 0.05% and 0.1%) for Cu, B, and Mo, four concentrations (0.05%, 0.1%, 0.15% and 0.2%) for Zn, and three concentrations (0.2%, 0.5% and 1%) for Mn were prepared for the experiment. Three repeats were made for each concentration. The temperature for germination was controlled at 22℃.

Topdressing (spraying on foliage): Topdressing was

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conducted at the nursery by spraying trace element solution on foliage. Two concentrations was prepared for each kind of trace elements as Cu: 0.02%, 0.05; B: 0.02%, 0.05; Mn: 0.2%, 0.5%; Mo: 0.02%, 0.05%; Zn: 0.05%, 0.1%.

Each concentration treatment repeats three times. The area of each pretreatment and contrast is 0.5 m^2. An isolated zone was kept between different treatments. Four treating methods were designed as follows:

Method A: Soaking seeds in trace elements solution before sowing; Method B: Soaking seeds in distilled water and spraying trace element solution on foliage (Topdressing) during the seedling stage; Method C: soaking seeds in trace element solution, and spaying trace element solution during seedling stage; Method D: Sowing seeds after only soaking with distilled water as contrast

During the whole process of experiment, the medicines such as chemical fertilizer, herbicide and pesticide were not used, and the other tending methods were the same as those of the general nursery management.

Result and analysis

Seed germination

The germination rates of the seeds treated with Mn and Zn trace elements had a relative remarkable increase, 7%~14% higher than the control group, and showed an increasing trend with the increase of concentration. The seeds treated by boron element presented a low germination rate, which was close to or lower than that of control group (See Table 3). In addition, the experimental results also showed that the activity of peroxide enzymes of the seeds treated by all the experimental trace elements, except boron, with varied concentrations, was higher then the control (Table 4). Seeds treated with Mn and Zn elements showed an increase trend in enzyme activity with the increase of the concentration, which is consistent with the variation of germination rate.

Table 3. The germination rate of the *Pinus tabulaeformis* seeds treated with different trace elements

Elements	Concentration (%)	Germination rate (%)		Absolute germinationrate (%)		Germination potential (%)	
		Mean value	Relative value	Mean value	Relative value	Mean value	Relative value
Cu	0.02	68.04	103	87.75	106	40.73	121
	0.05	68.00	103	88.73	107	38.00	113
	0.1	60.34	*	78.73	*	34.84	103
B	0.02	60.34	*	79.32	*	34.84	103
	0.05	65.99	100	76.59	*	39.62	118
	0.1	65.93	100	85.22	103	41.64	124
Mo	0.02	71.33	108	87.38	106	42.67	127
	0.05	74.50	113	85.93	104	41.00	122
	0.1	75.33	114	85.50	104	53.67	160
Mn	0.2	71.33	108	93.48	113	42.84	127
	0.5	74.50	113	92.10	114	45.84	135
	1	75.33	114	91.56	115	48.74	145
Zn	0.05	70.67	107	87.45	106	35.50	106
	0.1	70.0	107	88.71	107	38.67	114
	0.15	71.67	109	90.18	109	42.36	126
	0.2	74.67	113	92.60	112	44.00	131
Control	-	65.99	100	82.73	100	33.62	100

* The blank represents that the value is less than the value of control.

Topdressing

Topdressing was conducted by spraying trace element fertilizer on foliage separately on August 8, 18 and September 3, 2000 and investigation was carried out on September 10, 2000. The status of seed growth after topdressing was shown in Table 5. The trace elements Mn and Mo produced good effect on seedling growth. Base on this result, we continued to spray the two elements on the seedlings on May 20 and 27, 2001, and investigated the treated seedlings on June 4, 2001 (Table 6). The results showed that, the effect of Mn and Mo trace elements on seedling growth declined when making continuous application in second year. For example, the height of the seedling treated with Mo element increased 24%~54% in the year 2000 but only 5%~18% in 2001 compared to the control. The seedlings with topdressing grew better than those only treated with seed soaking.

Table 4. The peroxide enzyme activity of *Pinus tabulaeformis* seeds treated with different trace elements

Elements	Concentration	Activity of peroxide enzyme of seed
Cu	0.02	0.082
	0.05	0.041
	0.01	0.08
B	0.02	0.01
	0.05	0.01
	0.01	0.016
Mo	0.02	0.059
	0.05	0.026
	0.01	0.042
Mn	0.2	0.029
	0.5	0.032
	1.0	0.042
Zn	0.05	0.03
	1.0	0.014
	0.15	0.047
	0.2	0.048
Control	0.25	

Table 5. The quality of the foliage application with fertilizer on *Pinus tabulaeformis* seedling

Treating method	elements	Concentration (%)	Height (cm)		Root length (m)		Collar-diameter (mm)		Fresh weight (g)				Dry weight (g)			
			Mean value	%	Mean value	%	Mean value	%	Above ground		Below ground		Above ground		Below ground	
Method A	Cu	0.02	5	118	17.4	136	1.35	150	8.54	123	2.14	150	2.4	123	0.7	110
		0.05	4.75	112	17	133	1.11	123	8.7	125	2.9	202	2.5	130	0.66	110
	B	0.02	4.4	104	13.8	108	0.75	-	8.4	122	2.6	172	2.1	109	0.69	107
		0.05	4.75	112	12		1.25	139	7.8	112	2.1	146	1.8	-	0.6	100
	Mn	0.2	4.65	106	12.5		0.95	106	6.8	-	1.9	151	2.1	109	0.69	107
		0.5	5.25	124	12.5		0.95	106	8.4	121	2	140	2.5	130	0.62	100
	Mo	0.02	6.55	154	15.5	122	0.85	-	9.3	134	2.8	196	2.3	121	0.6	100
		0.05	5.25	124	12.5		0.95	106	8.4	121	2	140	2.5	130	0.62	100
	Zn	0.05	4.6	103	16.5	132	0.3		8.1	117	2.7	191	1.98	103	0.53	
		0.1	4.2		14.7	115	0.85		6.4		1.9	132	1.7		0.6	100
Method B	Cu	0.02	4.4	104	13.5	106	1.1	128	8.4	121	2.5	173	1.73		0.51	
		0.05	5	118	12.3		1	111	7.3	105	1.7	118	2.18	114	0.62	108
	B	0.02	4		15.5	122	1.1	122	8.7	125	2.7	189	2.3	120	0.63	103
		0.05	4.3	105	13.3	104	1.2	133	2.9	114	2.4	167	2.33	121	0.6	100
	Mn	0.2	5.5	128	14.5	114	0.9	100	8.7	125	2.45	171	2.5	130	0.76	127
		0.5	6.3	148	16	103	0.9	100	3.7	125	2.39	164	2.4	126	0.82	137
	Mo	0.02	5.5	132	13.3	104	0.95	106	11.0	159	2.45	171	2.9	150	0.66	110
		0.05	5	118	14.5	105	1.05	111	9.33	134	1.8	129	2.8	146	0.67	112
	Zn	0.05	4.6	107	13.4	105	1.1	122	7.14	104	1.7	124	2.3	120	0.63	103
		0.1	5	118	14	110	0.9	100	6.8		1.6	117	1.54		0.53	
Method C	Cu	0.02	4.25	100	14	110	1.15	128	8.07	116	1.74	122	2.3	120	0.61	102
		0.05	4.15		13.4	105	0.85		5.02		2.37	166	2.18	114	0.59	
	B	0.02	6.1	144	13.6	106	0.9	100	8.64	124	1.55	108	1.58	-	0.72	120
		0.05	5.75	133	13.8	108	1.95	106	7.5	107	2.14	170	2.9	151	0.68	113
	Mn	0.2	5.4	127	14.8	116	1.0	111	8.9	125	2.14	150	2.67	139	0.6	100
		0.5	6.45	152	13.5	106	0.9	100	9.9	135	92.7	109	2.42	126	0.63	103
	Mo	0.02	6.75	159	13.5	106	0.95	106	8.9	129	0.5	143	1.95	102	0.6	100
		0.05	5.6	132	1.5	118	0.9	100	8.3	134	2.4	166	2.5	130	0.6	172
	Zn	0.05	5	106	14.2	177	0.85		5.5		1.97	133	1.95	102	0.56	
		0.1	5	118	12.5		0.7		1.5	100	2.09	145	2.36	123	0.61	102
Control			4.25	100	12.2	100	0.9	100	6.94	100	1.43	100	1.92	100	0.6	100

Note: Method A: Soaking seeds in trace elements solution before sowing; Method B: Soaking seeds in distilled water and spraying trace element solution on foliage (Topdressing) during the seedling stage; Method C: soaking seeds in trace element solution and spaying trace element solution on foliage during seedling stage

Table 6. Growth status of *Pinus tabulaeformis* seedlings treated by Mn and Mo trace elements

Treating method	element	Concentration (%)	Height (cm)	Relative value	Root length /cm	Relative value	Collar diameter /mm	Relative value	Fresh weight /g	Relative value
Method A	Mn	0.2	8.24	106	21.22		1.67		17.3	
		0.5	8.37	108	21.99		1.66		19.2	102
	Mo	0.02	9.14	118	21.37		1.81	104	25	132
		0.05	8.18	105	18.79		1.75	101	18.4	
Method B	Mn	0.2	11.04	142	24.72	110	2.03	117	26.5	140
		0.5	10.26	132	24.21	109	1.94	111	23.1	122
	Mo	0.02	12.13	156	23.70	106	2.31	133	28.5	151
		0.05	10.3	133	23.45	105	1.87	107	22.8	121
Method C	Mn	0.2	9.46	122	23.34	105	1.75	101	21.8	115
		0.5	9.52	123	20.70	-	1.7	-	19.3	102
	Mo	0.02	11.39	147	23.56	106	1.89	109	28.4	150
		0.05	10.15	131	21.34	-	1.84	106	25.8	137
Control			7.77	100	22.29	100	1.74	100	18.9	100

Table 7. The chlorophyll content of the *Pinus tabulaeformis* seedling treated by Mn and Mo element in 2001

Method	Element	Concentration (%)	Chlorophyll a	Chlorophyll b	Total mean value	Total relative value(%)
Method A	Mn	0.2	1.064	0.401	1.464	106
		0.5	0.961	0.130	1.091	
	Mo	0.02	1.115	0.212	1.327	144
		0.05	0.990	0.285	1.275	
Method B	Mn	0.2	1.964	0.277	2.241	162
		0.5	1.352	0.534	1.886	137
	Mo	0.02	1.868	0.426	2.294	166
		0.05	1.521	0.472	1.993	145
Method C	Mn	0.2	1.901	0.340	2.241	163
		0.5	1.752	0.268	2.020	147
	Mo	0.02	1.745	0.522	2.267	165
		0.05	1.620	0.484	2.104	153
Control			1.058	0.320	1.378	100

Discussions

The experiments of seed germination indicated that the seeds treated by Mn element of 1% and 0.2% concentrations had best germination result, and the germination rates were raised by 9%~19% and 12%~14% compared with the untreated ones. This is attributed to the fact that Mn is the structural component of chlorophyll, which has effect on the oxidation-deoxidization in plant, and the necessary element of many enzyme systems. The germination rate of the seeds treated by boron element is the lowest (Table 3). This is because boron element impacts the activity of enzyme in the process of the carbohydrate metabolizability and inhibits the enzymes that activate the seed germination. Still the effect of boron element of higher or lower concentration on seed germination needs further study.

The treating method of spraying trace element solution on seedlings for topdressing has better result on growth of seedlings in contrast to that of soaking seeds in trace element solution. Soaking seed may have promoting effect on seedling growth in certain degree in the first year, but from the second year the chlorophyll content shows a decrease trend (Table 7), and its effect gradually decrease.

Topdressing with trace elements in low concentration could produce good result. For example, both the height and chlorophyll content of the seedlings treated by the topdressing of Mn and Mo element were much higher than that of the control group. This is closely related to the function of Mn and Mo trace element in plant. Mn element is the structural component of the chlorophyll, the application of fertilizer contained Mn element promotes the composing of the chlorophyll and activates the enzyme. Mo element is the component of nitrogenase and it can help plant sufficiently utilize nitrogen. However, topdressing with high concentration of Mn and Mo will inhibit the physiological activity of seedling.

Besides concentration and spraying technique, many other factors such as the rainfall during application, illumination, and the phase of growth as well as the fertile of soil will also have influences on the result of trace element application. Thus further study needs to be carried out in future.

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